

IN THE SPECIFICATION

Please amend the title as follows:

~~BINARY BAC VECTOR AND USES THEREOF~~

METHOD OF INTRODUCING

HETEROLOGOUS DNA INTO A NON-PLANT HOST CELL

IN THE CLAIMS:

1. (currently amended): A method of introducing heterologous DNA into a non-plant host cell thereby producing a gene product in said cell, said method comprising:
 - a) inserting heterologous DNA encoding said gene product into a unique restriction endonuclease cleavage site of a vector, said vector comprising:
 - i) a backbone which includes a first origin of replication capable of maintaining heterologous DNA as a single copy in *Escherichia coli* host cell, and which further includes a second origin of replication capable of maintaining heterologous DNA as a single copy in an *Agrobacterium tumefaciens* host cell;
 - ii) a unique restriction endonuclease cleavage site for insertion of heterologous DNA; and
 - iii) left and right *Agrobacterium* T-DNA border sequences flanking said unique restriction endonuclease cleavage site, said left and right T-DNA border sequences allowing introduction of heterologous DNA located between left and right T-DNA border sequences into a non-plant cell;
 - b) transforming a non-plant cell so as to introduce said heterologous DNA into said cell; and
 - c) expressing said heterologous DNA in said non-plant cell so as to produce the gene product encoded by said heterologous DNA into said cell, wherein said host cell is a yeast cell, a filamentous fungi or a mammalian cell.

2. (canceled)
3. (currently amended): The method of claim 2 1, wherein the yeast cell is *Saccharomyces cerevisiae* or *Kluyveromyces lactis*.
4. (currently amended): The method of claim 2 1, wherein the filamentous fungus is from the genus *Aspergillus*.
5. (currently amended): A method of producing a gene product in a non-plant host cell, said method comprising:
 - a) inserting heterologous DNA encoding said gene product into a unique restriction endonuclease cleavage site of a vector, said vector comprising:
 - i) a backbone which includes a first origin of replication capable of maintaining heterologous DNA as a single copy in *Escherichia coli* host cell;
 - ii) a unique restriction endonuclease cleavage site for insertion of heterologous DNA; and
 - iii) left and right *Agrobacterium* T-DNA border sequences flanking said unique restriction endonuclease cleavage site, said left and right T-DNA border sequences allowing introduction of heterologous DNA located between left and right T-DNA border sequences into a non-plant host cell;
 - b) introducing the resulting vector into said non-plant host cell; and

- c) expressing said heterologous DNA in said non-plant host cell so as to produce the gene product encoded by said heterologous DNA, wherein said non-plant host cell is a yeast cell, filamentous fungi or mammalian cell.
6. (original): The method of claim 5, wherein said vector further includes a second origin of replication capable of maintaining heterologous DNA as a single copy in an *Agrobacterium tumefaciens* host cell.
7. (canceled)
8. (canceled)
9. (canceled)
10. (canceled)
11. (original): The method of claim 1 or 5, wherein the heterologous DNA is obtained from genomic DNA of prokaryotic cells.
12. (original): The method of claim 1 or 5, wherein the heterologous DNA is obtained from genomic DNA of eukaryotic cells.
13. (original): The method of claim 1 or 5, wherein said first origin of replication comprises an F origin from *Escherichia coli*.
14. (original): The method of claim 1 or 5, wherein said second origin of replication comprises an Ri origin from *Agrobacterium rhizogenes*.

15. (original): The method of claim 6, wherein first origin of replication comprises an F origin from *Escherichia coli* and said second origin of replication comprises an Ri origin from *Agrobacterium rhizogenes*.
16. (original): The method of claim 1 or 5, wherein said unique restriction endonuclease cleavage site comprises a BamHI cleavage site.
17. (canceled)
18. (original): The method of claim 1 or 5, further comprising a selection marker for incorporation of heterologous DNA into said vector.
19. (canceled)
20. (original): The method of claim 1 or 5, further comprising a selection marker for introduction of said heterologous DNA into *Escherichia coli*.
21. (canceled)
22. (original): The method of claim 6, further comprising a selection marker for the introduction of said heterologous DNA into *Agrobacterium tumefaciens*.
- ~~21~~23. (currently amended): The method of claim 1 or 5, further comprising a selection marker for introduction of said heterologous DNA into a non-plant eukaryotic cell, said selection marker located between said left and right T-DNA border sequences.
- ~~22~~24. (currently amended): The method of claim 1 or 5, wherein said selection marker is located adjacent to said left T-DNA border sequence.

~~23~~25. (currently amended): The method of claim 21, wherein said kanamycin resistance gene comprises a GUS-NPTII gene.

~~24~~26. (currently amended): The method of claim 20, wherein said selection marker comprises a hygromycin resistance gene.

~~25~~27. (currently amended): The method of claim 1 or 5, wherein said backbone further comprises an origin of conjugal transfer.

~~26~~28. (currently amended): The method of claim ~~25~~ 27, wherein said origin of conjugal transfer comprises an oriT origin from plasmid RK2.

~~27~~29. (currently amended): A non-plant eukaryotic host cell containing a vector, said vector comprising:

- a) a backbone which includes a first origin of replication capable of maintaining heterologous DNA as a single copy in *Escherichia coli* host cell;
- b) a unique restriction endonuclease cleavage site for insertion of heterologous DNA; and
- c) left and right *Agrobacterium* T-DNA border sequences flanking said unique restriction endonuclease cleavage site, said left and right T-DNA border sequences allowing introduction of heterologous DNA located between left and right T-DNA border sequences into a non-plant host cell;
- d) a heterologous DNA inserted at said unique restriction endonuclease cleavage site; and

- e) a second origin of replication capable of maintaining heterologous DNA as a single copy in an *Agrobacterium tumefaciens* host cell.

~~28~~30. (currently amended): The non-plant eukaryotic host cell of claim ~~27~~ 29, wherein the host cell is a yeast cell.

~~29~~31. (currently amended): The non-plant eukaryotic host cell of claim ~~27~~ 29, wherein the host cell is a mammalian cell.

~~30~~32. (currently amended): The host cell of claim ~~27~~ 29, wherein the heterologous DNA is from a eukaryotic cell.

~~31~~33. (currently amended): The host cell of claim ~~27~~ 29, wherein the heterologous DNA is from a prokaryotic cell.

~~32~~34. (currently amended): A method of isolating a DNA encoding a desired gene product from a genomic library of DNA comprising:

- a) inserting heterologous DNA from a genomic library of DNA into a vector, said vector comprising:

- i) a backbone which includes a first origin of replication capable of maintaining heterologous DNA as a single copy in *Escherichia coli* host cell;
- ii) a unique restriction endonuclease cleavage site for insertion of heterologous DNA; and
- iii) left and right *Agrobacterium* T-DNA border sequences flanking said unique restriction endonuclease cleavage site, said left and right T-DNA

border sequences allowing introduction of heterologous DNA located
between left and right T-DNA border sequences into a non-plant host cell;

- b) introducing the resulting vector, into said non-plant host cell; and
- c) expressing said heterologous DNA in said non-plant host cell so as to produce the gene product encoded by said heterologous DNA,
- d) screening the cultured host cells for those cells that express the desired gene product, and
- e) isolating the DNA encoding the desired gene product from those cells that express the desired gene product.

3335. (currently amended): The method of claim 32 34, wherein said vector further includes a second origin of replication capable of maintaining heterologous DNA as a single copy in an *Agrobacterium tumefaciens* host cell.

3436. (currently amended): The method of claim 32 34, wherein the host cell is *Escherichia coli*.

3537. (currently amended): The method of claim 32 34, wherein the host cell is a non-plant eukaryotic cell.

3638. (currently amended): The method of claim 32 34, wherein the non-plant eukaryotic host cell is a yeast cell.

3739. (currently amended): The method of claim 32 34, wherein the non-plant eukaryotic host cell is a mammalian cell.

~~38~~40. (currently amended): The host cell of claim ~~32~~ 34, wherein the genomic library is obtained from prokaryotic cells.

~~39~~41. (currently amended): The host cell of claim ~~32~~ 34, wherein the genomic library is obtained from eukaryotic cells.

REMARKS/ARGUMENTS

Applicants have amended the title as requested by the Examiner. In addition, Applicants have amended the claims to further define the present invention. No new matter has been added by virtue of the amendments to the claims.

Applicants respectfully submit that the amendments to claims 5 and 19 have obviated the claim rejections, which should therefore be withdrawn.

Claims 1 – 41 stand rejected under 35 U.S.C. § 112, second paragraph.

With regard to the rejection of claims 1, 5, 29 and 34, Applicants respectfully submit that the term “heterologous DNA” is defined in the specification as “DNA not normally present in the particular host cell transformed by the vector”. *See*, paragraph 19. Accordingly, Applicants respectfully submit that in light of the specification, it is clear what the term means.

With regard to the remainder of the rejections, applicants respectfully submit that the cancellation of the claims have obviated this rejection, which should therefore be withdrawn.

Claims 1, 5 – 7, 11 and 13 – 28 stand rejected under 35 U.S.C. § 102(b) as being anticipated by Hamilton (1997).

Applicants respectfully disagree and request that this rejection be withdrawn for the following reasons.

Applicants submit that claims 1 and 5 have been amended to recite that the host cell is a yeast cell, filamentous fungus, or a mammalian cell. Applicants respectfully submit that such host cells are not disclosed in Hamilton (1997), and thus the rejection should be withdrawn.

Claims 1, 5 – 7, 11 and 13 – 28 stand rejected under 35 U.S.C. § 102(b) as being anticipated by Hamilton et al. (1999).

Applicants respectfully disagree and request that this rejection be withdrawn for the following reasons.

Applicants submit that claims 1 and 5 now recite that the host cell is a yeast cell, filamentous fungi, or mammalian cell. Applicants further submit that Hamilton et al. (1999) do not teach such host cells, and thus the rejection should be withdrawn.

Claims 1, 5 – 8, 10, 11, 13 – 29, 31, 34 and 39 stand rejected under 35 U.S.C. § 103 as being unpatentable over Hamilton (1997) in view of Hamilton (U.S. Patent No. 5,733,744).

Applicants respectfully disagree and request that this rejection be withdrawn.

As noted above, Hamilton (1997) does not teach yeast, filamentous fungi or mammalian host cells. Applicants respectfully submit that the secondary reference, Hamilton '744 does not make up for the difference. Although the '744 patent might teach the advantages of BIBAC vectors, those advantages are taught for **plant** cell transformation. There is no teaching or suggestion to transform host cells other than plants. Without such suggestion, (and without hindsight) the skilled artisan would have no motivation to use the BIBAC vector to transform host cells other than plant cells. Accordingly, Applicants respectfully request that this rejection be withdrawn.

Claims 1 – 9, 11 -15, 17 – 30, 32 – 38, 40 and 41 stand rejected under 35 U.S.C. § 103(a) as being unpatentable over Hamilton (1997) in view of Hamilton ('744), de Groot et al. (1998), and Bundock et al. (1995).

Applicants respectfully disagree and request that this rejection be withdrawn for the following reasons.

As admitted by the Examiner, Hamilton (1997) does not teach a method of introducing a heterologous DNA into a non-plant host cell that is a yeast cell, a filamentous fungi cell, *S.cervisiae*, *K.lactis*, or *aspergillus*.

Applicants respectfully submit that the secondary references do not make up for the deficiencies of Hamilton (1997).

The '744 patent is again directed to the use of BIBAC vectors for **plant** cell transformation.

de Groot et al. is directed to agrobacterium-mediated transformation of filamentous fungi. While Bundock et al. is directed to agrobacterium-mediated transformation of yeast.

Applicants respectfully submit that these references would in no way motivate one of ordinary skill in the art to use the BIBAC vector to transform host cells other than plant host cells. Further, the Examiner has provided no indication as to how one of ordinary skill in the art, without knowledge of the claimed invention, would piece together the teachings of four references to result in the claimed invention. Applicants submit that the only way to arrive at the present invention using the cited references is through impermissible hindsight obviousness. Consequently, Applicants submit that this rejection should be withdrawn.

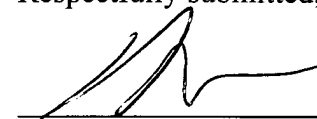
Applicants believe that the present application, as amended, is in a condition for allowance and issuance of the Patent is earnestly solicited.

Authorization is hereby given to the Commissioner to charge any deficient fees or to credit any overpayment to account no. 50-0850.

Date: 9/24/03

Customer No.: 26770

Respectfully submitted,



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